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A new *Tarentola* subspecies (Reptilia: Gekkonidae) endemic to Tunisia

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Abstract. Mitochondrial DNA sequences as well as morphological characters reveal that geckos of the genus *Tarentola* from Libya and central Tunisia are a monophyletic group which is different from both *T. mauritanica* and *T. deserti*. Consequently, we elevate the former subspecies *T. mauritanica fascicularis* to species rank. Together with *T. neglecta* and *T. mindiae*, *T. fascicularis* constitutes the sister group of *T. deserti*. *Tarentola fascicularis* comprises several evolutionary units, one of which we describe here as a subspecies endemic to south-central Tunisia.

Key words. *Tarentola*, gecko, Tunisia, Libya, North Africa, taxonomy.

INTRODUCTION

The Mediterranean geckos of the genus *Tarentola* Gray, 1825 are classified in the nominative subgenus *Tarentola*. The Canary Islands are inhabited by geckos of the subgenera *Tarentola* (Eastern Canaries) and *Makariogecko* (western Canaries, Selvagens and Cape Verde Islands) whereas other subgenera inhabit sub-Saharan Africa and the Caribbean (Joger 1984a, b). To date, the nominative subgenus comprises the following species: *Tarentola mauritanica* (L., 1758), *T. deserti* Boulenger, 1891, *T. angustimentalis* Steindachner, 1891, *T. boehmei* Joger, 1984. Although no subspecies have been described in the latter three species, the North African populations of *T. mauritanica* have been assigned to a number of subspecies; including, *T. m. mauritanica*, *T. m. fascicularis* (Daudin, 1802) from Egypt, *T. m. juliae* Joger, 1984, and *T. m. pallida* Geniez et al., 1999, both from Morocco. *Tarentola angustimentalis* (eastern Canary Islands) and *T. deserti* (northern margin of the Sahara Desert) used to be classified as subspecies of *T. mauritanica* until they were elevated to species rank (Joger 1984b). In the case of *T. deserti*, sympatric records of *T. deserti* and *T. mauritanica* in Tunisia and Algeria gave reason to assume that these were separate biological species.

Tarentola deserti and *T. mauritanica* can be distinguished from each other in a number of characters (Table 1, Figs 1 and 2). In the field, *T. deserti* is characterized often by its very large size, pale, often rosy body colour and a yellowish or ochre brown coloured iris, whereas typical *T. mauritanica* is smaller and has a grey body and iris colouration.

In contrast, in south-central Tunisia (Bou Hedma National Park and areas to the north of the Chott al Djerid salt pan) populations of *Tarentola* were found that show a mixture of characters of both species (Table 1, Fig. 3). Their size is smaller than *T. deserti*, but body and eye colour are close to *T. deserti* (Joger & Bischoff 1989; Joger 2003). A preliminary study of morphological and electrophoretic characters (Willand 1997; Joger et al. 1998) showed that these geckos cluster morphologically with *T. mauritanica* (yet not with any of its described subspecies), whereas their dorsal colour and pattern is close to *T. deserti* and their blood plasma protein alleles are distinct and not shared by neither *T. mauritanica* nor *T. deserti*.

Previous molecular genetic studies (Carranza et al. 2002; Rato et al. 2010) of North African *Tarentola* were biased in that they concentrated on Moroccan populations but largely neglected Tunisian and Libyan populations. In this study, we use both morphological and molecular samples of Tunisian and Libyan *Tarentola* to determine the affinities and clarify the taxonomy of the enigmatic *Tarentola* of south-central Tunisia.

MATERIALS AND METHODS

Animals were collected during several trips to Morocco, Algeria, Tunisia (U.J.) and Libya (I.B.). Specimens from Egypt were kindly provided by Sherif Baha El Din and Adel Ibrahim. Blood samples were taken by heart puncture or from muscle tissue of dead animals and preserved

Fig. 1. *Tarentola mauritanica* (Tunisia).

Operational taxonomic units (OTUs) were defined using a combination of mitochondrial DNA clades and geographic proximity. Linear Discriminant Function Analyses (LDFA) were used to find variables that separate the groups. Principal Component Analyses (PCA) were applied to see whether groups are distinguishable without previous definition of OTUs. Significance of character differences were tested with t-tests.

DNA was extracted from the preserved samples using standard procedures. Universal primers were used to amplify mitochondrial 12S rRNA (372 bp) and 16s rRNA (2 fragments of 448 and 604 bp). Sequences were determined using an automated sequencer, and aligned with CLUSTAL-W omitting gaps. Sequences from Genbank were added in some cases. A sequence of a paratype of the new subspecies was submitted to GenBank (IB47:

Table 1. Distinguishing characters of North African *Tarentola* (mean ± standard deviation). Significant differences from *T.* sp. (Tunisia) are highlighted (in bold). Significance values are given for males (first value) and females separately (if different). *** P<0.001; ** P<0.01; * P<0.05; n.s. not significant.

characters	*T.mauritanica* - Tunisia N=22	*T.*sp.- Tunisia N=15	*T.*sp.-complex-Libya N=139	*T.deserti* ssp.- Libya N=73	*T.d.deserti* - N-Africa N=24
Maximal head + body length	83.5	86	79	77.7	103.3
Lamellae under 1st toe	11.0 ± 1.0	**10.3 ± 0.8**	**10.9 ± 0.6 n.s./***	**11.0 ± 0.7 n.s./***	**12.7 ± 1.1 *** **
Lamellae under 4th toe	**16.8 ± 1.0** ***	**15.3 ± 1.0**	15.8 ± 1.0	**15.9 ± 0.8 n.s./****	**18.2 ± 1.3 *** **
Lamellae under 5th toe	20.5 ± 1.5	**19.7 ± 1.1**	20.2 ± 1.6	20.3 ± 1.1	**22.8 ± 1.3 *** **
Ventral scales	37.7 ± 3.3	**34.3 ± 2.7**	36.6 ± 3.2	**40.0 ± 2.7** ***	**40.1 ± 2.0** ***
Dorsal tubercles	13.5 ± 1.0	**12.1 ± 0.5**	12.8 ±1.4	**13.4 ± 0.9** **/***	12.2 ± 0.7
Gular scales	41.1 ± 4.7	**43.3 ± 6.6**	43.3 ± 4.7	45.3 ± 5.4	**56.0 ± 6.9** ***
Interorbitals	**14.9 ± 1.2** **	**13.7 ± 1.0**	**14.9 ± 0.7** ***	**14.6 ± 0.9** **	14.5 ±0.7
Infralabials	8.0 ± 0.7	**7.9 ± 0.7**	7.9 ± 0.5	8.0 ± 0.5	**8.8 ± 0.6** ***
Supralabials	7.8 ± 0.6	7.7 ± 0.7	7.9 ± 0.5	8.0 ± 0.4	8.0 ± 0.4
Relative inter-orbital width	0.38 ± 0.03 0.39 ± 0.02	**0.42 ± 0.04** **0.41 ± 0.03**	**0.36 ± 0.04** *** 0.37 ± 0.04 n.s.	**0.36 ± 0.04** *** **0.37 ± 0.05** *	0.43 ± 0.06 0.42 ± 0.04
Relative ear to mental distance	0.84 ± 0.03 0.88 ± 0.06	**0.91 ± 0.03** **0.90 ± 0.03**	**0.66 ± 0.07** *** **0.69 ± 0.21** **	**0.66 ± 0.06** *** **0.68 ± 0.09** ***	0.89 ± 0.04 0.88 ± 0.03

in 96% ethanol. Voucher specimens were preserved in 80% ethanol.

Morphological data were taken as described by Joger (1984a). Measurements were taken to the nearest 0.1 mm and normalized as proportion to body length. Pholidotic counts were taken unaltered.

HQ437282). Phylogenetic trees were constructed using a Bayesian Markov Chain Monte Carlo inference applying an evolution model suggested by MODELTEST 3.7. Statistical support for branches was indicated by posterior probability values (MrBayes).

Fig. 2. *Tarentola deserti* (Biskra, Algeria).

Fig. 3. *Tarentola* sp. (Bou Hedma, Tunisia).

RESULTS

Morphological comparisons

CDF plots (Figs 4a, b) show that *T.* sp. (Tunisia) are different morphologically from *T. deserti* as well as from undescribed *Tarentola* from western Libya ('*T.* sp. complex'). When North African populations of *T. mauritanica* are compared with *T.* sp. (Tunisia), only males appear distinct, whereas females cluster with *T. mauritanica* from Tunisia and Morocco (Fig. 5b).

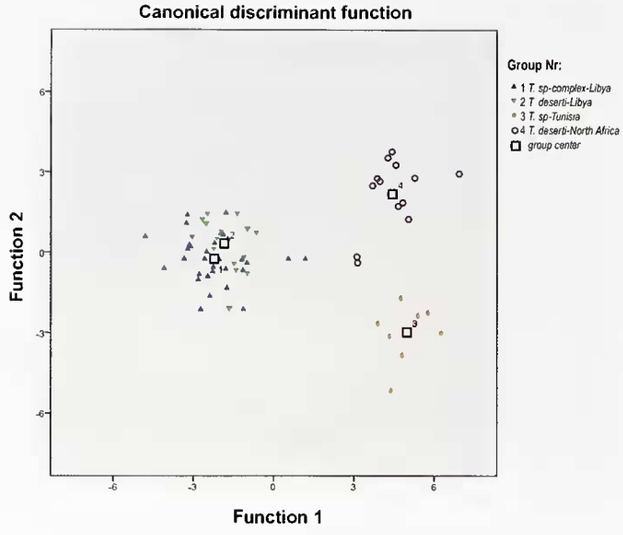
Significantly different characters which distinguish Tunisian *Tarentola* from Libyan and other North African populations are shown in Table 1.

Molecular genetic affinities

Mitochondrial gene sequences (12S rRNA, 16s rRNA) reveal that *T.* sp. (Tunisia) does not cluster with *T. mauritanica* but with undescribed Libyan *Tarentola* (Fig. 8). These Libyan populations form several geographically restricted, monophyletic clades; the most western ones are sister to *T.* sp. (Tunisia) – yet with rather low statistical support. *Tarentola deserti*, *T. mauritanica fascicularis* (Egypt, East Libya), and even *T. neglecta* appear more closely related to the western Libyan-Tunisian group of clades than *T. m. mauritanica*. This is highly supported statistically (1.00 posterior probability). It is noteworthy that the populations near the (neo-) type locality of *T. m.*

Table 2. Uncorrected "P" distance between main clades, estimated of evolutionary divergence between sequences, based on pairwise analysis of 1433 bp mtDNA sequences.

	Clade A	Clade B	Clade C	Clade D	Clade E	Clade F	Clade G	Clade H
Clade A								
Clade B	0.0921							
Clade C	0.0972	0.0704						
Clade D	0.1180	0.0977	0.0692					
Clade E	0.1052	0.0853	0.0562	0.0705				
Clade F	0.0974	0.0634	0.0630	0.0802	0.0645			
Clade G	0.1145	0.0873	0.0662	0.0665	0.0694	0.0510		
Clade H	0.1141	0.0886	0.0691	0.0711	0.0706	0.0578	0.0270	
Outgroup	0.1234	0.1407	0.1357	0.1545	0.1564	0.1422	0.1531	0.1490



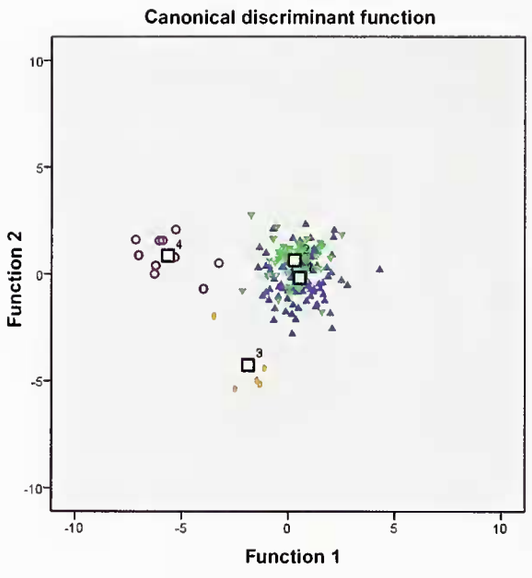


Fig. 4. CDF plots for *Tarentola* sp-Tunisia, *Tarentola deserti*-North Africa, *Tarentola sp*-complex-Libya (western Libyan *T. fascicularis*), and *Tarentola deserti*-Libya; males (above) and females (below).

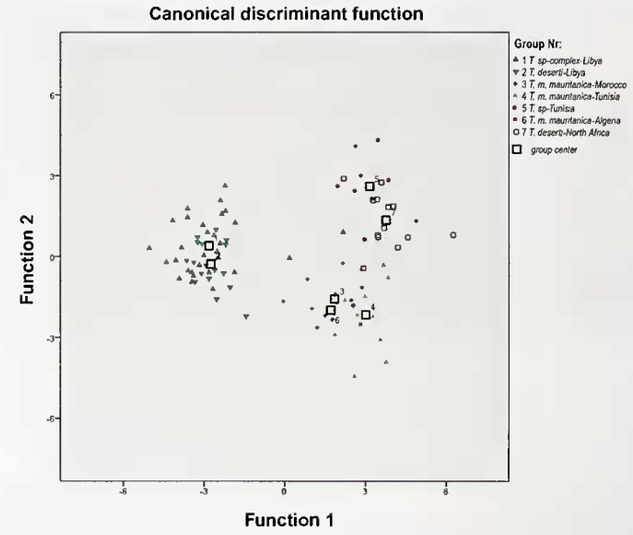


Fig. 5. CDF plots of different populations of North African *Tarentola*, males (above) and females (below) plotted separately.

fascicularis, in East Libyan Cyrenaica, belong to a separate clade (clade D).

Genetic distances among Libyan and Tunisian *Tarentola* clades are provided in Table 2.

Two of the mitochondrial clades occur sympatrically or parapatrically: *T. deserti* (Libya) and '*T.* sp. complex' (clade H) at Itwellia (western Libya), clades C and D along the desert road Tobruk-Ajdabiya in Cyrenaica.

DISCUSSION

Parapatric or sympatric occurrence of mitochondrial clades could be interpreted in different ways: either different biological species or co-existence of two mitochondrial distinct populations in a mixed interbreeding organismal population. In the case of *T. deserti* in NW Libya, there is evidence of the former explanation, as the molecular differences coincide with morphological differences.

Table 3. Variation of the Paratypes (part).

Variable	Mean	Minimum	Maximum	Standard deviation
Lamellae under 1st toe	10.27	9.00	12.00	0.80
Lamellae under 4th toe	15.27	14.00	17.00	0.96
Lamellae under 5th toe	19.67	18.00	21.00	1.11
Ventral scales	34.33	28.00	38.00	2.72
Supraorbital scales	5.73	5.00	6.00	0.46
Dorsal tubercles	12.13	12.00	14.00	0.52
Gular scales	44.21	32.00	55.00	5.82
Interorbital scales	13.73	12.00	15.00	0.96
Head length	17.18	13.89	21.03	2.16
Head+body length	56.08	44.12	72.33	7.62
Infralabialia	7.93	7.00	9.00	0.70
Supralabialia	7.73	6.00	9.00	0.70
Relative hindleg length	0.49	0.45	0.51	0.02
Relative head width	0.70	0.62	0.82	0.05
Relative head length	0.31	0.29	0.32	0.01
Relative foreleg length	0.36	0.32	0.40	0.02
Relative ear-snout length	0.90	0.84	0.95	0.03

When a conservative two-species concept (*T. mauritanica* – *T. deserti*) is applied, the mitochondrial tree unambiguously affiliates all sequenced Libyan OTUs with *T. deserti*, and not with *T. mauritanica*. The mitochondrial genetic distance between the central Tunisian *Tarentola*, the Libyan clades and *T. deserti* are lower than between the Tunisian clade and *T. m. mauritanica*. This supports the view that despite some morphological similarity, the Tunisian and Libyan-Egyptian clades are not subspecies of *T. mauritanica*.

Assigning these OTUs to *T. deserti* would, however, create a paraphyletic *T. deserti*, with *T. neglecta* and *T. mindiae* – which are without doubt separate species – within *T. deserti*. The most parsimonious taxonomic solution with regard to the cladogram is to subsume clades C, D, E, G, and H under one separate species.

Morphological data indicate that several of the mitochondrially defined populations, in particular if they occur in desert areas (Sabha in South Libya, Algerian and Tunisian but not Libyan *deserti*, and also a subclade of clade D) can be distinguished by larger size and higher scale counts. These size-linked characters may be locally favoured by environmentally triggered selection. On the other hand, morphological differences do not preclude genetic closeness, and genetically distant clades may share morphological similarity.

In conclusion, the genetically studied *Tarentola* from Libya and Egypt, as well as those from south central Tunisian, should be assigned to *T. fascicularis* (a former subspecies of *T. mauritanica*). Elevation of *fascicularis* to species rank is largely consistent with data of Rato et al. (2010), who distinguished two basal divisions in the subgenus *Tarentola*. One of these branches lead to *T. deserti* and *T. boehmei*, another to *T. angustimentalis* and *T. mauritanica* from Europe, Morocco, Algeria and northern Tunisia on one side, and to *T.* (*m.*) *fascicularis* and *Tarentola* from Lampedusa and Conigli Islands on the other side. Single individuals of '*T. mauritanica*' from Algeria and of '*T. deserti*' from Morocco were loosely connected to the latter clade, but we do not know which taxa were really represented by those samples. The Tunisian samples used by these authors clustered with *T. m. mauritanica*, but they were exclusively from northwestern Tunisia. True *T. mauritanica* exist in coastal areas of Tunisia and western Libya.

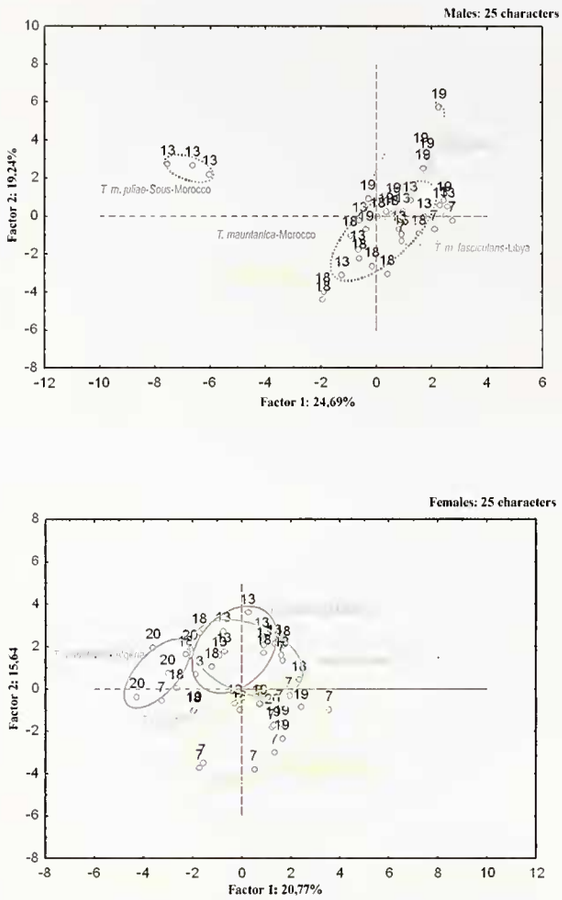


Fig. 6. PCA of *Tarentola* sp. (Tunisia), *T. m. fascicularis* (Libya), and different populations of *T. mauritanica*. Males above, females below.

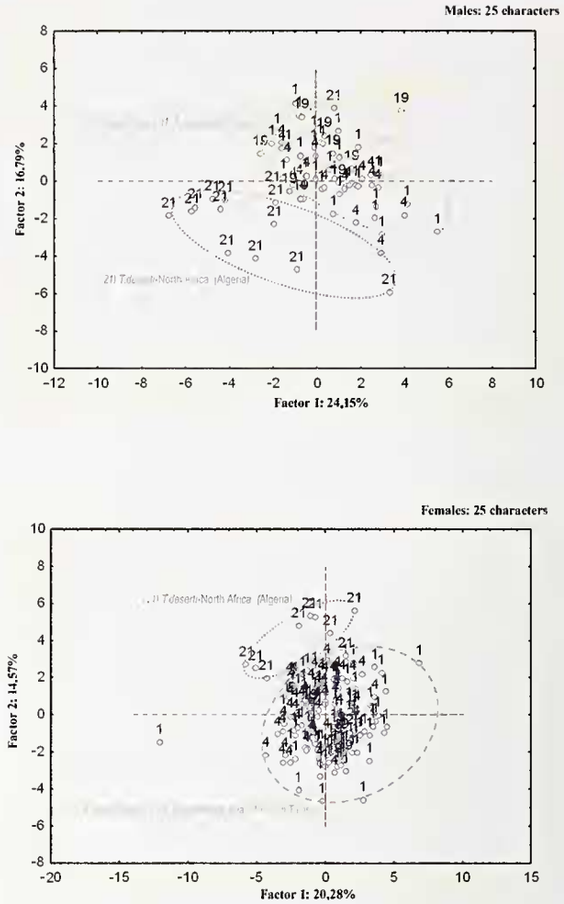


Fig. 7. PCA of *Tarentola deserti* (Libyan populations), *T.* sp. (western population of *T. fascicularis*, Libya) and *T.* sp. (Tunisia). Males above, females below.

In Tunisia, *T. deserti* exists in the extreme south (south-southeast of the Chott al Djerid) and a new subspecies of *T. fascicularis* in south central Tunisia between the Chott al Djerid and Djebel Bou Hedma. The description of this new subspecies is presented below.

Description of a new subspecies of *Tarentola fascicularis*

Tarentola fascicularis n. comb.

Gecko fascicularis Daudin, 1802

Tarentola mauritanica mauritanica, Loveridge 1947 (partim, non Linnaeus 1768)

Tarentola mauritanica fascicularis, Joger 1984

Terra typica (after designation of a neotype by Joger [1984]): Ain Teyanah, 20 km south of Benghazi, Libya.

Tarentola fascicularis wolfgangi ssp. n.

Holotype. State Natural History Museum Braunschweig (SNHM-BS) N 41980, male, collected 19 August 1998 by Ulrich Joger (Fig. 9).

Terra typica. Bou Hedma National Park, Tunisia (34.24'N, 9.23'E).

Paratypes. 33 specimens; SNHM-BS 39920-39930, 41981, Bou Hedma; HLMD 2105-2109, 2265-2271, 2363-2366, Bou Hedma; HLMD 1238-1240, Djebel Orbata/El Guettar; ZFMK 49525, 49526, Djebel Orbata: El Guettar.

Description of holotype. Measurements (mm). Head + body 61.0, tail 71.4, head length 19.8, head width 14.7, head height 10.8, interorbital width 8.4, distance snout-ear 17.7; foreleg 23.9, hindleg 30.2, distance between foreleg and hindleg 25.1, 4th toe length 5.1, toe width 2.0, diameter of eye 4.5.

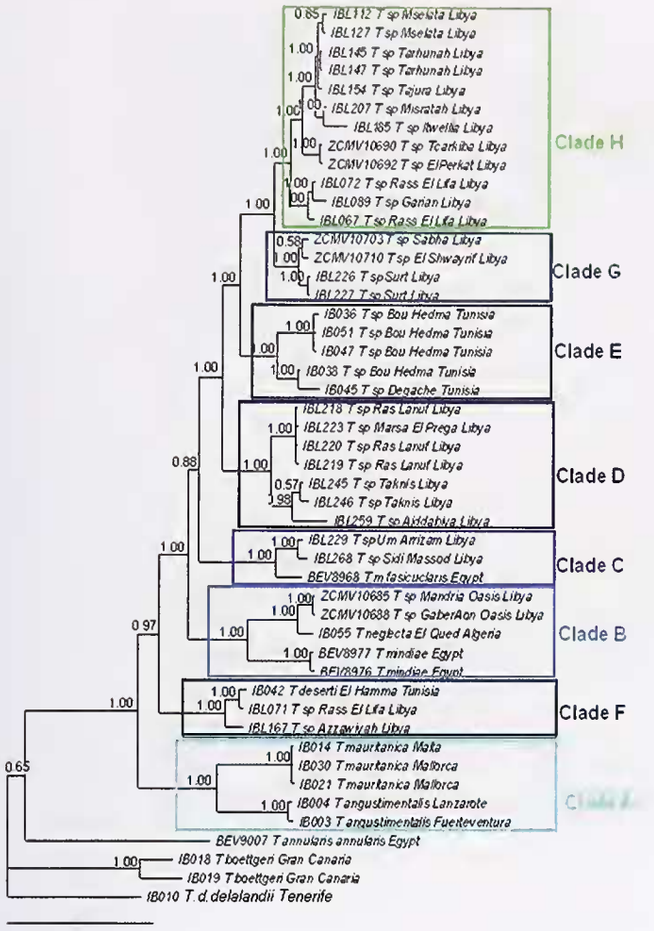


Fig. 8. 50% majority-rule consensus tree obtained from Bayesian MCMC analysis, based on 1433 bp mtDNA sequences, depicting the relationships among haplotypes. *Tarentola delalandii* designated as outgroup and Bayesian posterior probability values are given near branches.

Pholidosis. 36 longitudinal rows of ventral scales; 12 longitudinal rows of dorsal tubercles, bearing strong central keel from which barely visible keels derive laterally; 13 lamellae under 1st toe, 15 lamellae under 4th toe, 20 lamellae under 5th toe; 15 interorbital scales, 6 supraorbital scales; gular scales separated from mental by 3 scales, gular scale count 43; 10 supralabials, 7/8 infralabials; rostral divided, touching nostril; nasal scales separated by one scale proximally and one scale distally. Colour (in ethanol) whitish, without any visible pattern.

Fig. 9. Holotype of *Tarentola fascicularis wolfgangi* ssp. n.

Variability of paratypes. Colour (in ethanol) light or medium grey dorsally, whitish ventrally. Most specimens bear following pattern: dark line on side of head from eye to above ear. Paired dark spots, followed posteriorly by unpaired whitish spot (without clear margins) distributed on mid-dorsum as follows: one in front of shoulder, one behind shoulder, two on back, one on pelvic region, one on base of tail, followed by 9–10 unpaired half-rings around dorsal part of tail. Scale count variation is shown in Table 3.

Diagnosis. A small subspecies of *T. fascicularis*; maximum recorded body+head length in males 72.3 mm, in females 57.5 mm (up to more than 100 mm in male *T. deserti*, 81 mm in female *T. deserti*; in eastern Libyan *T. fascicularis*, 97 mm can be attained in males of the Ras Lanuf population, yet only 79 mm in *T. fascicularis* ssp. from northwestern Libya).

Tail length usually clearly longer than body+head (index body+head/tail 0.77-1.00; mean 0.84, as opposed to 0.98 in *T. deserti*, 0.96 in *T. f. fascicularis*, and 0.96 in *T. m. mauritanica*). Snout (ear openings to mental) significantly longer than in T. *fascicularis* and *T. deserti* subspecies from Eastern Libya (about 90% of head length as opposed to 60–70%).

Dorsal tubercles in 11–14 (most often 12) longitudinal rows, most often simply keeled (multiply keeled in *T. f. fascicularis*). 19–46 gular scales (45–59 gular scales in *T. d. deserti*). Different from all other Tunisian or Libyan *Tarentola* (except *T. neglecta* group) by lower number of ventral scales (34.3 +/- 2.7) and lower number of lamellae underneath 1st and 4th toes (1st 10.3 +/- 0.8, 4th 15.3 +/- 1.0). 15–22 scale rows or lamellae underneath 5th toes (16-21 in *T. f. fascicularis*, 21–25 in *T. deserti*). Different from Tunisian *T. mauritanica* by lower number of interorbital scales (13.7 +/- 1.0 versus 14.9 +/- 1.2); from Libyan *T. mauritanica* by lower number of sublabials (7.9 +/- 0.7 versus 8.7 +/- 0.7); from western Libyan subspecies of *T. fascicularis* by lower number of interorbital scales (13.7 +/- 1.0 versus 14.9 +/- 0.7). Rostral usually separated from nostril by small scales (in *T. f. fascicularis* rostral usually in contact with nostril).

Dorsal colour in life similar to *T. deserti*: rosy or yellowish, with yellowish iris (grey in *T. m. mauritanica*). Five dark transverse bands across back, often reduced to paired spots.

Distribution. Endemic to Central Tunisia; known from Gafsa (Djebel Orbata) in the West to Bou Hedma in the East, south to Degache and Tozeur at northern banks of Chott al Djérid.

Fig. 10. Terra typica of *Tarentola fascicularis wolfgangi* ssp. n., Djebel Bou Hedma, Tunisia.

Habitat. Bou Hedma National Park is famous for its relict subtropical savanna with *Acacia tortilis raddiana* as the dominating tree. The climate is semi-arid, with variable amounts of rainfall (annual mean about 250 mm) in autumn and winter. On the pediments of Djebel Bou Hedma (Fig. 10) as well as on the slopes of the mountain chains to the west and south of it, *Tarentola fascicularis wolfgangi* ssp. n. is found in rock crevices, on walls and underneath of road bridges; the geckos are active at night.

Derivatio nominis. The species is dedicated to Wolfgang Böhme on the occasion of his retirement as the most successful German curator of herpetology after Robert Mertens. The senior author feels, however, also a strong affinity to the other Wolfgang, Wolfgang Bischoff, who retired this year, too. His field companionship in North Africa will be ever remembered.

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